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Metagenomics of human microbiome: beyond 16s rDNA

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Abstract

The gut microbiota presents a symbiotic relationship with the human host playing a beneficial role in human health. Since its establishment, the bacterial community is subjected to the influence of many different factors that shape its composition within each individual. However, an important convergence is observed at functional level in the gut microbiota. A metatranscriptomic study of healthy individuals showed homogeneity in the composition of the active microbiota that increased further at functional level.

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The gut microbiota as an epigenetic landscape

The human gastrointestinal tract is the natural habitat for a large microbial community including species from Archaea, Bacteria, Virus and Eukaryota. Most of these microbes are symbiotic to the human host and beneficial to human health because of their contributions in nutrient processing, development of the immune system, colonization resistance and stimulation of a variety of other host activities [1,2].

The microbiota can be regarded as an essential 'organ' of the human body responsible for metabolic functions that human cells might not be able to carry out themselves. Concepts of evolutionary developmental biology [3] may be helpful to understand this ecosystem. In fact, little is known about the epigenetic landscape—following Waddington's metaphor—of this 'organ', in contrast to the human counterparts which are formed through differentiation of mother cells [4]. It is believed that the establishment of the microbiota in the intestine starts even before birth and that this ecosystem develops quickly over the first year of life [5,6]. A number of factors may influence this development. The specific species that colonize the gut, the niches they occupy, time, space, potential perturbations, interactions within the community and with the host as well as other factors related to the unique environment that each human being represents, shape up the taxonomic composition of the microbiota. This may explain the high compositional variability observed between subjects. However, these multiple configurations harbouring a vast number of genes, and hence potential functionality, may in practice be 'phenocopies' [7], in the sense of being capable of carrying out the same functions for the host, even though the metabolic pathways for those functions vary from subject to subject depending on the species involved. Also, the huge number of available genes, which may or may not express depending on the environmental conditions, provides this 'organ' with the ability to quickly adapt and react to environmental stresses and sudden changes, thus making it extremely robust from a functional perspective. We have tried to represent these concepts schematically and metaphorically in Fig. 1.

The next section provides some evidence in support of the epigenetic landscape approach mentioned above.

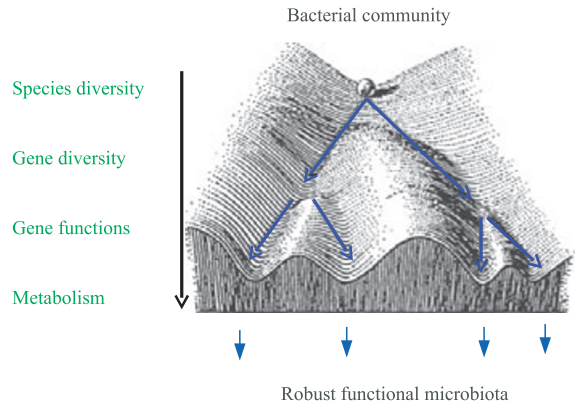

FIG. 1. Schematic representation of the microbiota's functional landscape.

Metatranscriptome of gut bacterial communities

Although major efforts have been devoted to describing the structure of the microbial populations in the human gut, very little is known about the activity of their members [8–11]. Some effort needs to be done to identify gene expression patterns *in vivo* so they could be used as makers for the description of bacterial physiology in the host. There is a necessity in the study of the global transcription of metagenomes to qualitatively and quantitatively establish a functional profile under a healthy status that can serve as a reference for comparison against that in a disease status.

To establish a functional profile in healthy individuals, we analysed the metatranscriptome of intestinal microbiota using faecal samples from ten volunteers [12]. We applied large-scale pyrosequencing to the RNA communities: 16S rRNA transcripts (7250 reads per sample on average) as a marker of the structure of the active bacterial community and the mRNA fraction (1500 reads per sample on average) for the functions present in this habitat and the microorganisms involved. To study the taxonomic classification of the active microbiota in faecal samples, each read previously classified as a 16S transcript was BLAST searched against the Ribosomal Database Project II [13]. The correspondence analysis showed that the samples were relatively homogeneous in active bacterial composition. The human gut microbiome presents many physiological properties that are lacking in the host and can therefore be considered as essential for human life. To determine the potential functions of faecal microbiota in the samples, we performed a homology search by BLASTX against the gCOG database [12]. Fig. 2 shows a uniform functional pattern in all the samples from healthy individuals, the most abundant functional categories being those corresponding to the functions G (carbohydrate transport and metabolism), J (translation, ribosomal structure) and C (biogenesis and energy production and conversion). Additionally, we found that the distribution of bacterial families across functional categories is also rather uniform except for the categories of cell motility (N) and secondary metabolite biosynthesis, transport and catabolism (Q) (Fig. 3). Thus, the taxonomic composition of the active intestinal microbiota is fairly similar among individuals. This is in contrast to the much larger heterogeneity observed in the composition of the entire microbiota (whether active or not). Additionally, this homogeneity further increases at the functional level.

Conclusions

Metatranscriptomic analysis of the gut microbiota revealed a functional profile that is more similar between individuals

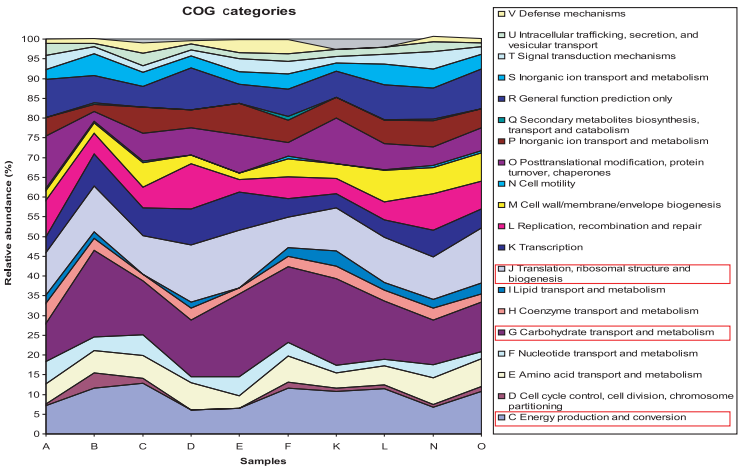

FIG. 2. Distribution of COG categories in the metatranscriptomes.

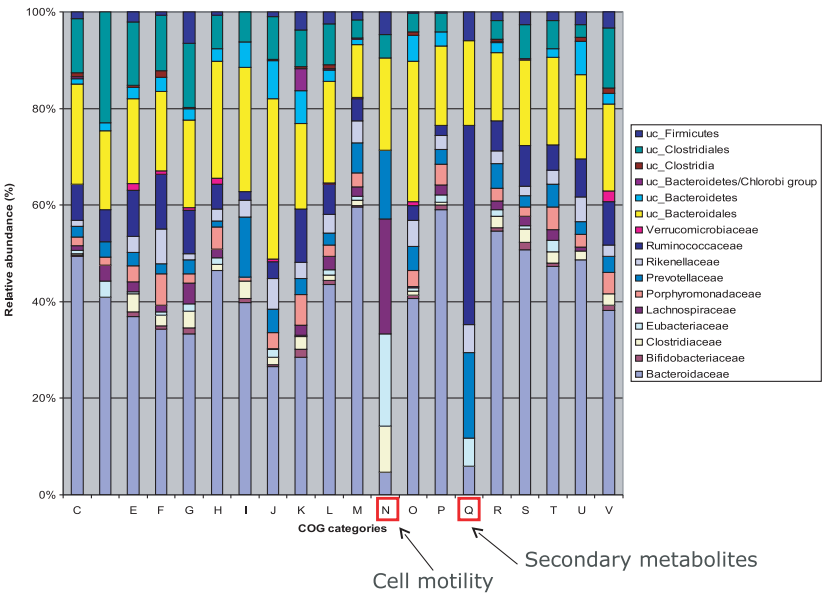

FIG. 3. Barplot of the distribution of taxa in functional categories.

than the taxonomic composition obtained from the metagenomic data. Thus, the evolutionary forces seem to drive the microbiome to a convergent functional pattern through different epigenetic landscapes.

Nevertheless, further research is needed to clarify the evolutionary development that leads to the different metabolic roles of the gut microbiota.

Transparency Declaration

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